

## Effects of Microplastics on Germination and Seedlings Growth of Wheat (*Triticum aestivum* L.)

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### Abstract

Microplastics pose a serious threat to both ecosystems and human health. It is increasingly detected in agricultural soil but how these microplastics affects on plant growth system that has remained poorly understood. The present investigation was assessed the effect of microplastics in wheat (*Triticum aestivum* L.) seedlings growth. Different microplastics in aqueous solution showed varied response on wheat seedlings growth. It was noted that after 10 days of seedlings growth in petri dishes, the WMRI-I wheat showed maximum efficiency of germination percentage (87.5%), shoot elongation (6.68 cm), root elongation (6.62 cm) and lateral root formation (4.38 roots/seedling) than WMRI-II and WMRI-III. At all the cases, the minimum growth response was noticed in WMRI-III wheat. The F-value reveals that shoot and root length were significantly different towards microplastic treatments. The present findings also indicate that PPLCW (microplastic plus polythene) treatment had more negative impact on wheat seedlings growth.

**Keywords:** Growth, Microplastics, Seedlings, Wheat.

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## Introduction

Microplastics are small pieces of plastic, less than 5 mm in length, that generate in both the aquatic and terrestrial environment as a consequence of plastic degradation. Microplastics are present in a variety of products, from cosmetics to synthetic clothing to plastic bags and bottles (Revel et al. 2018). Many of these products readily enter the environment as wastes. They can be either primary (manufactured for special purpose) or secondary (breakdown of larger plastic items into small fragments) forms. The degradation of plastic waste generates microplastic particles. Microplastics are now regarded as the most pollutant contaminants in the world that severely affects the whole ecosystem (Klaine et al. 2012). These particles are always detected as a mixture of various polymer types and shapes. However, we have limited knowledge about the effect of combined microplastics on plant-soil systems. Agricultural ecosystems are now polluted by microplastics in terrestrial environments due to different modern agricultural practices (Rillig and Lehmann 2020, Hofmann et al. 2023). The main path of microplastics entering the agricultural soil includes plastic mulch, plastic-coated fertilizers, biofertilizers, wastewater irrigation and others (Mahon et al. 2017). The application of plastic mulch and organic fertilizers likely increased microplastic contamination in agricultural soils, as plastic mulch can fragment over time and some organic fertilizers (e.g., composts or biosolids) may contain embedded plastic particles. Thus microplastic pollution in agro-ecosystem, is getting severely increased with changing soil biophysical properties and plant performances (Fei et al. 2020, Shen et al. 2022). Microplastics can influence plant growth, crop yield, nutrient utilization efficiency etc.

Wheat (*Triticum aestivum* L.) stands as the most important cereal crop throughout the world as a dietary staple food. In Bangladesh, it is the second most important crop after rice. The growth of wheat seedlings is influenced by soil characteristics like nitrogen and carbon contents. Soil, especially agricultural land, has emerged

as a significant reservoir of microplastics (Mahon et al. 2017), the majority of research efforts was concentrated on microplastics in aquatic ecosystems (Cole et al. 2011, Auta et al. 2017). Given the burgeoning concern of microplastic contamination in soil ecosystems and the prime importance of wheat as a food grain in Bangladesh, numerous fundamental research inquiries remain unanswered regarding the impacts of microplastic pollution on wheat. To the best of our knowledge, such investigations have not yet been conducted in Bangladesh, which is prompting our interest in undertaking this research endeavor.

## Materials and Methods

### Plant materials

Three popular wheat varieties (WMRI-I, WMRI-II and WMRI-III) were used to conduct the present investigation; these were collected from the Wheat and Maize Research Institute (WMRI), Rajshahi, Bangladesh.

### Preparation of microplastic containing aqueous solution

1. Three types of plastic materials (a. crude plastic particles, b. polythene c. a mixture of crude plastic plus polythene) were used. At first step, the crude particles were crushed by hammer into small pieces and passed through sieve (3-5 mm) which were considered as microplastic particles and kept in the beaker.
2. 10 g of each type of plastic materials were taken into 3 separate beakers.
3. Add 100 ml of boiled hot water separately in the above 3 beakers. The step was repeated by adding hot water in every beaker until 10 days to make the final volume of each beaker is 1000ml. The beakers were covered properly with aluminum foil and kept at lab conditions (temperature  $25\pm1^{\circ}\text{C}$  and light intensity of 800-1000 lux).
4. The mixtures were kept for 30 days and then filtered by cheese cloth. The filtrate was preserved as stock solutions of subjected microplastics and followed the subsequent experiment.

### Experimental design and treatments

The experiment was laid out in a Completely Randomized Design (CRD) with factorial arrangements having four treatments with three replicates. Each replicate consisted of one petri dish containing four seed samples. The four different microplastic treatments- i) Control (no microplastic traces), ii) PCW (plastic containing water), iii) PLCW (polythene containing water), and iv) PPLCW (plastic plus polythene containing water) were applied to three wheat varieties to assess their growth.

### Seedlings growth and data collection

The microplastic treatments were added daily to each petri dish in an amount sufficient to moisten the tested seeds. The control petri dish was moistened only with distilled water. The experiment was extended over a period of ten days where no further germination occurred. A seed was considered as germinated when radicle emerged and its length  $\geq 0.2$  cm. The data were recorded on germination percentages, shoot length, root length and number of lateral roots of wheat seedlings. The values of variable factors were evaluated by ANOVA at 5% level of significance.

## Results

### Germination percentages

The germination percentages of wheat seeds treated with different microplastic treatments were recorded at 3, 7 and 10 days. On average, the highest percentages of germination was recorded in WMRI-I (87.5%) followed by WMRI-II and WMRI-III. It was also noted that the microplastic treatments from PLCW-PPLCW showing lower germination percentages especially in WMRI-II and WMRI-III wheat. The efficiency of germination depends on variety and treatments (Table 1).

**Table 1:** Effect of microplastics in aqueous solution on germination percentages of wheat seeds after 3, 7 and 10 days of culture in petri dishes.

Wheat varieties	Microplastic treatments	Germination (%) in days		
		3	7	10
WMRI-I	Control	100	100	100
	PCW	100	75	75
	PLCW	75	100	100
	PPLCW	75	75	75
	Mean	87.5	87.5	87.5
WMRI-II	Control	100	100	100
	PCW	100	100	100
	PLCW	50	50	50
	PPLCW	75	75	75
	Mean	81.25	81.5	81.25
WMRI-III	Control	100	100	100
	PCW	75	75	75
	PLCW	75	75	75
	PPLCW	50	50	50
	Mean	75	75	75

PCW = Plastic containing water, PLCW = Polythene containing water, PPLCW = Plastic plus polythene containing water.

**Shoot length:** There was no significant difference in shoot length among the wheat varieties at 3, 7 and 10 days. The shoot length of wheat seedlings were increased rapidly with the increased days intervals. At the end period, WMRI-II achieved the highest shoot length of 9.4 cm at PCW treatment while WMRI-III had the lowest (2.5 cm) at PPLCW treatment. In general, the control treatment showed the maximum shoot length compared to the microplastic treatments except the WMRI-II wheat. At the end period (after 10 days), microplastic treatments showed significant differences in shoot elongation and revealed that it was more stressful than control in shoot elongation of wheat seedlings (Table 3).

**Root length:** The root growth is very important for the establishment of any seedlings. The wheat seedlings showed varied responses in root elongation with different microplastic treatments (Table 2). The root elongation was rapidly increased with the extended day's but the trend of elongation was gradually decreased from control to PPLCW treatments. The rooting performances were noted highest in WMRI-I after 10 days.

**Table 2:** Effect of microplastics in aqueous solution on seedlings growth of wheat after 3, 7 and 10 days of culture in petri dishes.

Wheat varieties	Microplastic treatments	Shoot length (cm)			Root length (cm)			No. of lateral roots		
		3 d	7 d	10 d	3 d	7 d	10 d	3 d	7 d	10 d
WMRI-I	Control	0.68	7.6	8.7	0.95	7.3	7.45	4.25	4.25	4.25
	PCW	1.05	2.6	4.36	1.15	3.8	5.4	4.25	4.25	4.25
	PLCW	1.15	5.9	8.5	2.03	6.5	7.03	4.75	4.75	4.75
	PPLCW	0.70	3.5	5.18	1.13	4.67	6.6	4.25	4.25	4.25
	Mean	0.89	4.9	6.68	1.32	5.57	6.62	4.38	4.38	4.38
WMRI-II	Control	0.95	5.02	6.25	0.83	5.3	5.65	4.25	4.25	4.25
	PCW	0.93	7.7	9.4	0.95	5.8	6.25	4.50	4.50	4.50
	PLCW	0.95	4.18	8.75	1.33	3.7	7.65	3.00	3.00	3.00
	PPLCW	0.98	3.9	4.2	1.40	2.4	3.53	3.25	3.25	3.25
	Mean	0.95	5.2	7.15	1.13	4.3	5.77	3.75	3.75	3.75
WMRI-III	Control	1.13	7.2	8.5	1.03	5.9	6.4	4.50	4.5	4.50
	PCW	0.68	3.48	5.43	1.50	3.02	4.5	3.50	3.5	3.50
	PLCW	0.83	3.75	5.9	1.88	3.78	5.47	3.50	3.5	3.50
	PPLCW	0.68	1.0	2.5	0.55	1.00	2.8	3.25	3.25	3.25
	Mean	0.83	3.85	5.58	1.24	3.43	4.85	3.69	3.69	3.69

PCW = Plastic containing water, PLCW = Polythene containing water, PPLCW = Plastic plus polythene containing water.

Among the microplastic treatments, PLCW showed better performance in root elongation, although the control treatment consistently exhibited the highest root elongation over the 10-day period. On varietal performances, rooting growth was noted highest in WMRI-I and the lowest was WMRI-III wheat. Table 3 shows that wheat varieties and the microplastic treatments both were significantly different in root elongation.

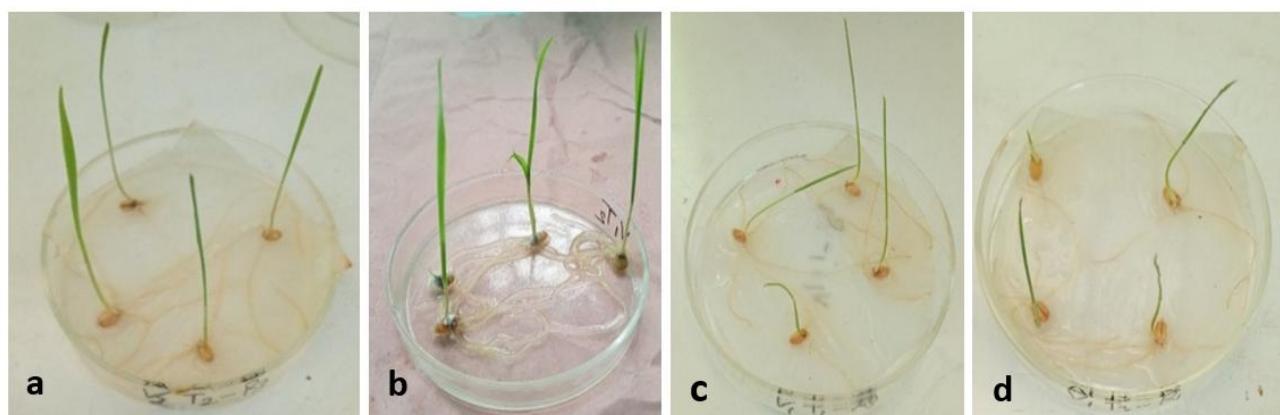
#### Number of lateral roots

The lateral root formation of wheat seedlings was not much influenced by microplastic treatments irrespective of day's intervals. The highest number of lateral root formation was noted in WMRI-I while the lowest was in WMRI-III wheat. The maximum lateral root formation was observed at control treatment. It was also noticed that microplastic treatments may not be stressful to the tested wheat varieties in lateral root initiation. ANOVA, revealed that there was no significant differences among the wheat varieties and the treatments, in lateral root formation of wheat seedlings (Table 3).

**Table 3:** Analysis of variance for shoot length, root length and number of lateral roots of wheat seedlings with different microplastic treatments in aqueous solution after 3, 7 and 10 days of culture in petri dishes.

Sources	df	F-value								
		After 3 days			After 7 days			After 10 days		
		Shoot length (cm)	Root length (cm)	No. of lateral roots	Shoot length (cm)	Root length (cm)	No. of lateral roots	Shoot length (cm)	Root length (cm)	No. of lateral roots
Varieties A)	2	0.33ns	0.14ns	0.98ns	2.29ns	4.06*	1.37ns	2.22ns	3.34*	1.66ns
Treat. (B)	3	0.11ns	1.51ns	0.57ns	1.76ns	5.34*	0.88ns	7.27*	4.84*	1.23ns
AB	6	0.51ns	0.49ns	0.48ns	1.60ns	1.35ns	0.89ns	4.23*	1.35ns	1.05ns
Error	36									

\*significant and ns, non-significant (ns) at 5% level.



**Fig. 1 a-d:** Seedlings growth of wheat at different microplastic treatments. (a) Control, (b) PCW (c) PLCW, and (d) PPLCW treatment.

#### Discussion

Microplastic effects on wheat seedlings growth were investigated. It was noted that microplastic with different treatments had diverse effects on germination and seedling growth of three different wheat varieties. During the growth stages of plants, seed germination is one of the prime steps which determine the success of a plant into the soil. At the initial stage of seed germination (after 3 days) with different microplastic treatments, WMRI-I wheat performed better in germination rate, root length, lateral root formation and shoot elongation. The

results also consisted for 7 days interval. At the 10-day period, a significant difference was noticed among the microplastic treatments in shoot and root elongations of wheat seedlings, but the varietal differences were not significant for shoot length and number of lateral root formation. These insignificant effect of microplastics have also been reported in other crops like onion (Giorgetti et al. 2020, Shi et al. 2022), Cucumber (Bouaicha et al. 2022) and rice (Spano et al. 2022). The root elongation of wheat seedlings was significantly affected by microplastic treatments irrespective of wheat varieties. Microplastics have been demonstrated to inhibit root growth in a variety of plants including wheat (Qi et al. 2018), broad beans (Jiang et al. 2019), and onions (Lian et al. 2020). The control treatment triggered the highest root length compared to other treatments. The shoot and root elongation was greatly affected by PPLCW treatment at all the wheat varieties.

## Conclusion

The effect of microplastics on wheat seedlings growth is complex and depends on multiple factors like microplastic type, size, concentration and the specific wheat variety. Different wheat varieties may exhibit varying responses to microplastic exposure. While some studies showing inhibition while others suggest promotion or no significant impact which is highlighting the need for further research to fully understand the long-term ecological implications of microplastic pollution on wheat seedlings growth both laboratory and field conditions.

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**Author's contribution:** MJT and MMU collect samples and data, conducted experiments, SKN prepare the draft manuscript, MHR designed the experiment, supervised the study and finally corrected the manuscript. All authors have read and approved the final manuscript.

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